

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the applications:

Listing of Claims:

Claims 1-124 (canceled)

125. (currently amended) A ~~recombinant protein encoded by~~ polynucleotide which comprises two nucleic acid subsequences and which encodes a TNF binding protein, wherein the first nucleic acid subsequence encodes a soluble fragment of an insoluble human TNF receptor protein, wherein said insoluble human TNF receptor protein has an apparent molecular weight of about 75 kilodaltons as determined on a non-reducing SDS-polyacrylamide gel, and wherein the second nucleic acid subsequence encodes all of the domains of the constant region of a human immunoglobulin heavy chain other than the first domain of said constant region, and wherein said TNF binding recombinant protein exhibits specific TNF binding activity.

126. (canceled)

127. (currently amended) The polynucleotide recombinant protein of claim 125 wherein said soluble fragment comprises the amino acid sequence LCAP (SEQ ID NO:12).

128. (currently amended) The polynucleotide recombinant protein of claim 127 wherein said soluble fragment comprises the amino acid sequence VFCT(SEQ ID NO:8).

129. (currently amended) The polynucleotide recombinant protein of claim 128 wherein said soluble fragment comprises the amino acid sequence LPAQVAFXPYAPEPGSTC (SEQ ID NO: 10).

130. (currently amended) The polynucleotide recombinant protein of claim 125, 128 or 129 wherein said human immunoglobulin heavy chain is an IgG heavy chain.

131. (canceled)

132. (currently amended) The polynucleotide recombinant protein of claim 130
[[131]] wherein the human immunoglobulin heavy chain IgG is IgG₁.

133. (currently amended) A recombinant protein encoded by a polynucleotide
which comprises two nucleic acid subsequences and which encodes a TNF binding protein,
wherein the first nucleic acid subsequence hybridizes to the nucleic acid sequence of
SEQ ID NO: 3 under conditions that discriminate between the nucleic acid sequence of SEQ
ID NO: 3 and the nucleic acid sequence of SEQ ID NO: 1, and

wherein the first nucleic acid subsequence encodes a soluble fragment of an insoluble
human TNF receptor protein,

wherein said insoluble human TNF receptor protein has an apparent molecular weight
of about 75 kilodaltons as determined on a non-reducing SDS-polyacrylamide gel,

wherein the second nucleic acid subsequence encodes all of the domains of the
constant region of a human immunoglobulin heavy chain other than the first domain of said
constant region, and

wherein said TNF binding recombinant protein exhibits specific TNF binding activity.

134. (currently amended) The polynucleotide recombinant protein of claim 133
wherein the first nucleic acid subsequence is obtainable by a method comprising a step of
hybridizing an oligonucleotide probe encoding the peptide of SEQ ID NO: 10
(LPAQVAFXPYAPEPGSTC) to a cDNA library made from HL60 cell extracts.

135. (currently amended) The polynucleotide recombinant protein of claim 133
wherein said soluble fragment comprises the amino acid sequence LCAP (SEQ ID NO 12).

136. (currently amended) The polynucleotide recombinant protein of claim 135
wherein said soluble fragment comprises the amino acid sequence VFCT (SEQ ID NO: 8).

137. (currently amended) The polynucleotide recombinant protein of claim 136 wherein said soluble fragment comprises a nucleic acid sequence encoding the amino acid sequence LPAQVAFXPYAPEPGSTC (SEQ ID NO: 10).

138. (currently amended) The polynucleotide recombinant protein of any one of claims 133, 136 or 137 wherein said human immunoglobulin heavy chain is an IgG heavy chain.

139. (canceled)

140. (currently amended) The polynucleotide recombinant protein of claim 138, [[139]] wherein the human immunoglobulin heavy chain IgG is IgG₁.

141. (currently amended) A recombinant protein encoded by a polynucleotide which comprises two nucleic acid subsequences and which encodes a TNF binding protein, wherein the first nucleic acid subsequence encodes a soluble fragment of an insoluble human TNF receptor protein and comprises a nucleic acid sequence encoding a fragment of SEQ ID NO: 4,

wherein the soluble fragment of SEQ ID NO:4 comprises the amino acid sequence LCAP,

wherein the second nucleic acid subsequence encodes all of the domains of the constant region of a human immunoglobulin heavy chain other than the first domain of said constant region, and

wherein said TNF binding recombinant protein exhibits specific TNF binding activity.

142. (currently amended) The polynucleotide recombinant protein of claim 141 wherein the soluble fragment further comprises the amino acid sequence VFCT (SEQ ID NO: 8).

143. (currently amended) The polynucleotide recombinant protein of claim 142 wherein the soluble fragment further comprises the amino acid sequence LPAQVAFXPYAPEPGSTC (SEQ ID NO: 10).

144. (currently amended) The polynucleotide recombinant protein of claim 141 wherein the first nucleic acid subsequence is obtainable by a method comprising a step of hybridizing an oligonucleotide probe encoding the peptide of SEQ ID NO: 10 (LPAQVAFXPYAPEPGSTC) to a cDNA library made from HL60 cell extracts.

145. (currently amended) The polynucleotide recombinant protein of any one of claims 141, 142 or 143 wherein said human immunoglobulin heavy chain is an IgG heavy chain.

146. (canceled)

147. (currently amended) The polynucleotide recombinant protein of claim 145 [[146]] wherein the human immunoglobulin heavy chain IgG is IgG₁.

148. (currently amended) The polynucleotide recombinant protein of claim 145 wherein the second nucleic acid subsequence consists essentially of the immunoglobulin-encoding DNA sequence of pCD4H_γ1 vector (deposited at Deutschen Sammlung von Mikroorganismen und Zellkulturen GmbH (DSM) in Braunschweig, FRG under No. DSM 5314) or of pCD4-H_γ3 vector (deposited at Deutschen Sammlung von Mikroorganismen und Zellkulturen GmbH (DSM) in Braunschweig, FRG under No. DSM 5523).

149. (currently amended) The polynucleotide recombinant protein of claim 148 wherein the second nucleic acid subsequence consists essentially of the immunoglobulin-encoding DNA sequences of pCD4-H_γ1 vector.

Claims 150-154 (canceled)

155. (new) The polynucleotide of claim 125, wherein the TNF binding protein consists of the soluble fragment and all the domains of the constant region of the human immunoglobulin heavy chain other than the first domain of the constant region.

156. (new) A vector comprising the polynucleotide of claim 125.

157. (new) A host cell comprising the polynucleotide of claim 125.
158. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of any one of claim 125, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
159. (new) The method of claim 158 wherein the host cell is a CHO cell.
160. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 128, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
161. (new) The method of claim 160 wherein the host cell is a CHO cell.
162. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 155, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
163. (new) The method of claim 162 wherein the host cell is a CHO cell.
164. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 132, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
165. (new) The method of claim 164 wherein the host cell is a CHO cell.
166. (new) The polynucleotide of claim 133, wherein the TNF binding protein consists of the soluble fragment and all the domains of the constant region of the human immunoglobulin heavy chain other than the first domain of the constant region.
167. (new) A vector comprising the polynucleotide of claim 133.

168. (new) A host cell comprising the polynucleotide of claim 133.
169. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 133, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
170. (new) The method of claim 169 wherein the host cell is a CHO cell.
171. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 136, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
172. (new) The method of claim 171 wherein the host cell is a CHO cell.
173. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 166, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
174. (new) The method of claim 173 wherein the host cell is a CHO cell.
175. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 140, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
176. (new) The method of claim 175 wherein the host cell is a CHO cell.
177. (new) The polynucleotide of claim 141, wherein the recombinant protein consists of the soluble fragment and all the domains of the constant region of the human immunoglobulin heavy chain other than the first domain of the constant region.
178. (new) A vector comprising the polynucleotide of claim 141.

179. (new) A host cell comprising the polynucleotide of claim 141.
180. (new) A host cell comprising the polynucleotide of claim 142.
181. (new) A host cell comprising the polynucleotide of claim 143.
182. (new) A host cell comprising the polynucleotide of claim 144.
183. (new) A host cell comprising the polynucleotide of claim 177.
184. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 141, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
185. (new) The method of claim 184 wherein the host cell is a CHO cell.
186. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 142, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
187. (new) The method of claim 186 wherein the host cell is a CHO cell.
188. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 143, and
 - (b) isolating expressed protein from the cell mass or the culture medium.
189. (new) The method of claim 188 wherein the host cell is a CHO cell.
190. (new) A method of producing a protein, comprising the steps of:
 - (a) culturing a host cell comprising the polynucleotide of claim 144, and
 - (b) isolating expressed protein from the cell mass or the culture medium.

191. (new) The method of claim 190 wherein the host cell is a CHO cell.

192. (new) A method of producing a protein, comprising the steps of:

- (a) culturing a host cell comprising the polynucleotide of claim 177, and
- (b) isolating expressed protein from the cell mass or the culture medium.

193. (new) The method of claim 192 wherein the host cell is a CHO cell.

194. (new) A method of producing a protein, comprising the steps of:

- (a) culturing a host cell comprising the polynucleotide of claim 147, and
- (b) isolating expressed protein from the cell mass or the culture medium.

195. (new) The method of claim 194 wherein the host cell is a CHO cell.

196. (new) A polynucleotide which encodes a TNF binding protein consisting of

- (a) a soluble fragment of an insoluble human TNF receptor protein, and
- (b) all of the domains of the constant region of a human IgG₁ immunoglobulin heavy chain, other than the first domain of said constant region,

wherein said insoluble human TNF receptor protein has an apparent molecular weight of about 75 kilodaltons as determined on a non-reducing SDS-polyacrylamide gel,

wherein said soluble fragment comprises the amino acid sequences LCAP and VFCT,

and

wherein said TNF binding protein exhibits specific TNF binding activity.

wherein said TNF binding protein exhibits specific TNF binding activity.

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wherein said TNF binding protein exhibits specific TNF binding activity.

wherein said TNF binding protein exhibits specific TNF binding activity.

197. (new) The polynucleotide of claim 196 wherein the soluble fragment further comprises the amino acid sequence LPAQVAFXPYAPEPGSTC (SEQ ID NO: 10).

198. (new) A vector comprising the polynucleotide of claim 196.

199. (new) A host cell comprising the polynucleotide of claim 196.

200. (new) A method of producing a protein, comprising the steps of:

- (a) culturing a host cell comprising the polynucleotide of claim 196, and
- (b) isolating expressed protein from the cell mass or the culture medium.

201. (new) The method of claim 200 wherein the host cell is a CHO cell.

202. (new) A method of producing a protein, comprising the steps of:

- (a) culturing a host cell comprising the polynucleotide of claim 197, and
- (b) isolating expressed protein from the cell mass or the culture medium.

203. (new) The method of claim 202 wherein the host cell is a CHO cell.